

The 3'-5' ero plus F → Y Mut. Project N \_\_\_\_\_  
 Book No. \_\_\_\_\_

31

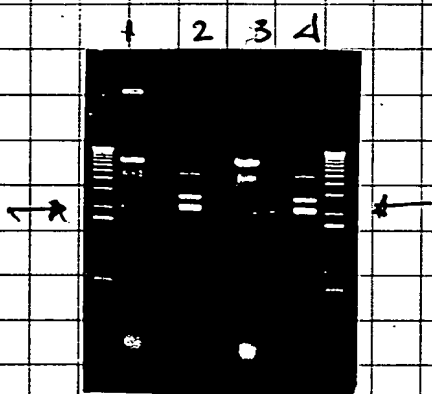
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chive Replace mutant fragment from RF to pUC19-Tne.

I cannot do it directly because there are two Sph I sites in pUC19-Tne.

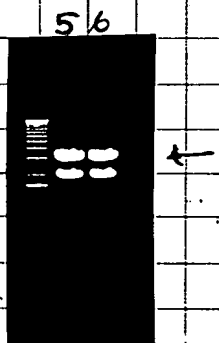
Therefore, first replace the mutant fragment in pUC19-Tne and then transfer into pUC19-Tne.

Digest pUC19-Tne with Sph I + Hind III. Digest M13mp19 mutant with Sph I + Hind III.



1 = RF 3'-5' ero - Sph I / Hind III  
 2 = pUC19-Tne "  
 3 = RF F → Y mutant "  
 4 = pUC19-Tne "

Purify only mutant fragment - to be ligated into pUC19-Tne Sph I / Hind III frag.



5 & 6 pUC19-Tne Sph I / Hind III

Purify the largest fragment. Ligate with the mutant fragments.

Transform DHOB.

Plate 10% & 90% →

Overnight at 30°C

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Read & Understood by me,

Date

Invented by

Date

11-11-95

6/12/95

Recorded by

Deb K. Khatun

5/1/95